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FILE COVERS 1907 - 9 May 2005  VOL 142 ISS 20
FILE LAST UPDATED: 8 May 2005  (20050508/ED)

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  This file contains CAS Registry Numbers for easy and accurate
  substance identification.

=> "HCV replicon"
          9089 "HCV"
            17 "HCVS"
          9093 "HCV"
                 ("HCV" OR "HCVS")
          3051 "REPLICON"
          1523 "REPLICONS"
          3755 "REPLICON"
                 ("REPLICON" OR "REPLICONS")
L1         154 "HCV REPLICON"
                 ("HCV"(W)"REPLICON")

=> "self replicating"
        313814 "SELF"
            23 "SELFS"
            38 "SELVES"
        313867 "SELF"
                 ("SELF" OR "SELFS" OR "SELVES")
          7719 "REPLICATING"
L2         604 "SELF REPLICATING"
                 ("SELF"(W)"REPLICATING")

=> L1 and L2
L3           4 L1 AND L2

=> "NS5" and l1
           837 "NS5"
L4          10 "NS5" AND L1

=> "5' non translation region"
       5851537 "5"
        717310 "NON"
            33 "NONS"
        717336 "NON"
                 ("NON" OR "NONS")
        345666 "TRANSLATION"
          1792 "TRANSLATIONS"
        347045 "TRANSLATION"
                 ("TRANSLATION" OR "TRANSLATIONS")
        891379 "REGION"
        412572 "REGIONS"
       1175575 "REGION"
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("REGION" OR "REGIONS")
L5 1 "5' NON TRANSLATION REGION"
("5"(W)"NON"(W)"TRANSLATION"(W)"REGION")

=> D L3 IBIB ABS 1-4

L3 ANSWER 1 OF 4 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2005:296055 CAPLUS
TITLE: Analysis of the 5' End Structure of HCV Subgenomic RNA Replicated in a Huh7 Cell Line
AUTHOR(S): Takahashi, Hitoshi; Yamaji, Masashi; Hosaka, Masahiro; Kishine, Hiroe; Hijikata, Makoto; Shimotohno, Kunitada
CORPORATE SOURCE: Institute for Virus Research, Kyoto University, Sakyo-ku, Kyoto, Japan
SOURCE: Intervirology (2005), 48(2-3), 104-111
CODEN: IVRYAK; ISSN: 0300-5526
PUBLISHER: S. Karger AG
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Objective: Recently, HCV subgenomic RNA that replicates in vitro in a certain cell line have been elucidated. Since the 5' end of the genome of pos. strand RNA viruses is often modified with a cap structure or a covalently linked protein, we have assessed structural feature of the HCV genome obtained from Huh7 cells in which HCV subgenomic RNA has been shown to efficiently self-replicate. Methods: HCV subgenomic RNA was obtained from the Huh7 and was analyzed for its 5' end. Results: Phosphorylation of the genomic RNA by polynucleotide kinase was observed only after treatment with phosphatase. The labeling efficiency of the genome with polynucleotide kinase was not enhanced by treatment with pyrophosphatase. Conclusion:It is suggested that the 5' end of HCV genomic RNA obtained from **HCV replicon** cells is not modified except phosphorylation. Furthermore, anal. of the 5' end of the HCV RNA obtained from the HCV subgenome **self-replicating** cells revealed the presence of two types of subgenomic RNA that contained either guanylate or adenylate at the 5' end. This result indicates that the 5' end of the subgenome in Huh7 cells is redundant and there is no significant evolutionary advantage between the two genomes.

L3 ANSWER 2 OF 4 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2004:905495 CAPLUS
DOCUMENT NUMBER: 141:375510
TITLE: Mammalian cell line comprising reporter-selectable hepatitis c virus replicon for screening antiviral agents
INVENTOR(S): Duggal, Rohit; Patick, Amy Karen; Zhang, Jie; Zhao, Weidong
PATENT ASSIGNEE(S): Agouron Pharmaceuticals, Inc., USA
SOURCE: U.S. Pat. Appl. Publ., 42 pp.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2004214178	A1	20041028	US 2003-422323	20030424
PRIORITY APPLN. INFO.:			US 2003-422323	20030424

AB The invention relates to a reporter-selectable hepatitis C virus (**HCV**) **replicon**, and use of the replicon to generate stable, human hepatoma cell lines. The replicon and cell lines are useful in the compound screening process in HCV drug discovery. The replicon comprises an HCV 5' NTR fused to a capsid coding region. The replicon also comprises a humanized Renilla luciferase (hRLuc) fused at its N-terminus said capsid and at its C-terminus to a foot and mouth disease virus (FMDV) 2A proteinase, which is fused to a neomycin phosphotransferase (NPTII) gene. The replicon further comprises an internal ribosome entry site (IRES) from an encephalomyocarditis virus

(EMCV), inserted downstream of the NPT II gene, which directs translation of HCV proteins NS3 to NS5B. The hRLuc2A-NPTII fusion, upon expression from the **HCV replicon**, is cleaved at the 2A peptide to generate the hRLuc protein and the subsequent hRLuc signal and the NPTII protein that confers resistance to G418. HCV NS3-5B polyprotein coding region contains adaptive mutations such as E1202G, T1208I and S2197P.

L3 ANSWER 3 OF 4 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2004:596260 CAPLUS
DOCUMENT NUMBER: 141:255367
TITLE: Evolution of naturally occurring 5' non-translated region variants of hepatitis C virus genotype 1b in selectable replicons
AUTHOR(S): Van Leeuwen, Hans C.; Reusken, Chantal B. E. M.; Roeten, Marko; Dalebout, Tim J.; Riezu-Boj, Jose Ignacio; Ruiz, Juan; Spaan, Willy J. M.
CORPORATE SOURCE: Department of Medical Microbiology, Center of Infectious Diseases, Leiden University Medical Center, Leiden, 2300 RC, Neth.
SOURCE: Journal of General Virology (2004), 85(7), 1859-1866
CODEN: JGVIAY; ISSN: 0022-1317
PUBLISHER: Society for General Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Quasispecies shifts are essential for the development of persistent hepatitis C virus (HCV) infection. Naturally occurring sequence variations in the 5' non-translated region (NTR) of the virus could lead to changes in protein expression levels, reflecting selective forces on the virus. The extreme 5' end of the virus' genome, containing signals essential for replication, is followed by an internal ribosomal entry site (IRES) essential for protein translation as well as replication. The 5' NTR is highly conserved and has a complex RNA secondary structure consisting of several stem-loops. This report analyses the quasispecies distribution of the 5' NTR of an HCV genotype 1b clin. isolate and found a number of sequences differing from the consensus sequence. The consensus sequence, as well as a major variant located in stem-loop IIIa of the IRES, was investigated using **self-replicating** HCV RNA mols. in human hepatoma cells. The stem-loop IIIa mutation, which is predicted to disrupt the stem structure, showed slightly lower translation efficiency but was severely impaired in the colony formation of selectable **HCV replicons**. Interestingly, during selection of colonies supporting autonomous replication, mutations emerged that restored the base pairing in the stem-loop. Recloning of these altered IRESs confirmed that these second site revertants were more efficient in colony formation. In conclusion, naturally occurring variants in the HCV 5' NTR can lead to changes in their replication ability. Furthermore, IRES quasispecies evolution was observed in vitro under the selective pressure of the replicon system.
REFERENCE COUNT: 34 THERE ARE 34 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L3 ANSWER 4 OF 4 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2002:504935 CAPLUS
DOCUMENT NUMBER: 137:74392
TITLE: **Self-replicating** RNA molecule from hepatitis C virus having adaptive mutations, and its uses in screening assay for HCV replication inhibitors
INVENTOR(S): Kukolj, George; Pause, Arnim
PATENT ASSIGNEE(S): Boehringer Ingelheim (Canada) Ltd., Can.
SOURCE: PCT Int. Appl., 140 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE

WO 2002052015	A2	20020704	WO 2001-CA1843		20011220
WO 2002052015	A3	20031120			

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

CA 2430607	AA	20020704	CA 2001-2430607		20011220
EP 1379660	A2	20040114	EP 2001-271930		20011220

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR

JP 2004516039	T2	20040603	JP 2002-553495		20011220
US 2002142350	A1	20021003	US 2001-29907		20011221
US 6706874	B2	20040316			
US 2003148348	A1	20030807	US 2002-309561		20021204
US 2004203020	A1	20041014	US 2003-686835		20031016
US 2004180333	A1	20040916	US 2004-789355		20040227
PRIORITY APPLN. INFO.:			US 2000-257857P	P	20001222
			WO 2001-CA1843	W	20011220
			US 2001-29907	A3	20011221
			US 2002-309561	A3	20021204

AB The present invention relates generally to a hepatitis C virus (HCV) RNA mol. that self-replicates in appropriate cell lines, particularly to a **self-replicating** HCV RNA construct having an enhanced efficiency of establishing cell culture replication. A unique HCV RNA mol. is provided having an enhanced efficiency of establishing cell culture replication. Novel adaptive mutations have been identified within the HCV non-structural region that improves the efficiency of establishing persistently replicating HCV RNA in cell culture. This **self-replicating** polynucleotide mol. contains, contrary to all previous reports, a 5'-NTR that can be either an A as an alternative to the G already disclosed and therefore provides an alternative to existing systems comprising a **self-replicating** HCV RNA mol. The G-->A mutation gives rise to HCV RNA mols. that, in conjunction with mutations in the HCV non-structural region, such as the G(2042)C/R mutations, possess greater efficiency of transduction and/or replication. The HCV RNA encoding polyprotein comprising one or more amino acid substitution selected from the group consisting of: R(1135)K; S(1148)G; S(1560)G; K(1691)R; L(1701)F; I(1984)V; T(1993)A; G(2042)C; G(2042)R; S(2404)P; L(2155)P; P(2166)L; M(2992)T; and E(1202)G is claimed. These RNA mols. when transfected in a cell line are useful for evaluating potential inhibitors of HCV replication.

=> D L4 IBIB ABS 1-10

L4 ANSWER 1 OF 10 CAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER:	2005:325014 CAPLUS
TITLE:	Development of cell-based assays for in vitro characterization of hepatitis C virus NS3/4A protease inhibitors
AUTHOR(S):	Chung, Victoria; Carroll, Anthony R.; Gray, Norman M.; Parry, Nigel R.; Thommes, Pia A.; Viner, K. Claire; D'Souza, Eric A.
CORPORATE SOURCE:	Department of Virology, GlaxoSmithKline Medicines Research Centre, Stevenage, SG1 2NY, UK
SOURCE:	Antimicrobial Agents and Chemotherapy (2005), 49(4), 1381-1390 CODEN: AMACCQ; ISSN: 0066-4804
PUBLISHER:	American Society for Microbiology
DOCUMENT TYPE:	Journal
LANGUAGE:	English

AB A recombinant vaccinia virus, expressing the NS3-to-**NS5** region

of the N clone of hepatitis C virus (HCV), was generated and utilized both in a gel-based assay and in an ELISA (ELISA) to evaluate the pyrrolidine-5,5-trans-lactams, a series of inhibitors of the HCV NS3/4A protease. The absolute levels of processed, mature HCV nonstructural proteins in this system were found to decrease in the presence of the trans-lactams. Monitoring of this reduction enabled end points and 50% inhibitory concns. to be calculated in order to rank the active compds. according to potency. These compds. had no effect on the transcription or translation of the NS3-5 polyprotein at concns. shown to inhibit NS3/4A protease, and they were shown to be specific inhibitors of this protease. The ELISA, originally developed using the vaccinia virus expression system, was modified to utilize Huh-7 cells containing an **HCV replicon**. Results with this assay correlated well with those obtained with the recombinant vaccinia virus assays. These results demonstrate the utility of these assays for the characterization of NS3/4A protease inhibitors. In addition, inhibitors of other viral targets, such as polymerase and helicase, can be evaluated in the context of the replicon ELISA.

REFERENCE COUNT: 35 THERE ARE 35 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 2 OF 10 CAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2005:233082 CAPLUS
DOCUMENT NUMBER: 142:348233
TITLE: Ribavirin resistance in hepatitis C virus replicon-containing cell lines conferred by changes in the cell line or mutations in the replicon RNA
AUTHOR(S): Pfeiffer, Julie K.; Kirkegaard, Karla
CORPORATE SOURCE: Department of Microbiology and Immunology, Stanford University School of Medicine, Stanford, CA, 94305, USA
SOURCE: Journal of Virology (2005), 79(4), 2346-2355
CODEN: JOVIAM; ISSN: 0022-538X
PUBLISHER: American Society for Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Ribavirin (RBV), used in combination with alpha interferon to treat hepatitis C virus (HCV) infections, is a guanosine nucleotide analog that can increase the error rate of viral RNA-dependent RNA polymerases, imbalance intracellular nucleotide pools, and cause toxicity in many cell types. To determine potential mechanisms of RBV resistance during HCV RNA replication, we passaged **HCV replicon**-containing cell lines in the presence of increasing concns. of RBV. RBV-resistant, **HCV replicon**-containing cell lines were generated, and the majority of RBV resistance was found to be conferred by changes in the cell lines. The resistant cell lines were defective in RBV import, as measured by [3H]RBV uptake expts. These cell lines displayed reduced RBV toxicity and reduced error accumulation during infection with poliovirus, whose replication is known to be sensitive to RBV-induced error. For one RBV-resistant isolate, two mutations in the replicon RNA contributed to the observed phenotype. Two responsible mutations resided in the C-terminal region of NS5A, G404S, and E442G and were each sufficient for low-level RBV resistance. Therefore, RBV resistance in **HCV replicon** cell lines can be conferred by changes in the cell line or by mutations in the **HCV replicon**.

REFERENCE COUNT: 57 THERE ARE 57 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 3 OF 10 CAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2004:792826 CAPLUS
DOCUMENT NUMBER: 142:128401
TITLE: Suppression of hepatitis C virus replicon by RNA interference directed against the NS3 and NS5B regions of the viral genome
AUTHOR(S): Takigawa, Yuki; Motoko, Nagano-Fujii; Deng, Lin; Hidajat, Rachmat; Tanaka, Motofumi; Mizuta, Hiroyuki; Hotta, Hak
CORPORATE SOURCE: Department of Microbiology, Kobe University Graduate

School of Medicine, Kobe, 650-0017, Japan
SOURCE: Microbiology and Immunology (2004), 48(8), 591-598
CODEN: MIIMDV; ISSN: 0385-5600
PUBLISHER: Center for Academic Publications Japan
DOCUMENT TYPE: Journal
LANGUAGE: English
AB RNA interference (RNAi) is a phenomenon in which small interfering RNA (siRNA), an RNA duplex 21 to 23 nucleotides (nt) long, or short hairpin RNA (shRNA) resembling siRNA, mediates degradation of the target RNA mol. in a sequence-specific manner. RNAi is now expected to be a useful therapeutic strategy for hepatitis C virus (HCV) infection. In the present study we compared the efficacy of a number of shRNAs directed against different target regions of the HCV genome, such as 5'-untranslated region (5'UTR) (nt 286 to 304), Core (nt 371 to 389), NS3-1 (nt 2052 to 2060), NS3-2 (nt 2104 to 2122), and NS5B (nt 7326 to 7344), all of which except for NS5B are conserved among most, if not all, HCV subtype 1b (HCV-1b) isolates in Japan. We utilized two methods to express shRNAs, one utilizing an expression plasmid (pAVU6+27) and the other utilizing a recombinant lentivirus harboring the pAVU6+27-derived expression cassette. Although 5'UTR has been considered to be the most suitable region for therapeutic siRNA and/or shRNA because of its extremely high degree of sequence conservation, we observed only a faint suppression of an HCV subgenomic replicon by shRNA against 5'UTR. In both plasmid- and lentivirus-mediated expression systems, shRNAs against NS3-1 and NS5B suppressed most efficiently the replication of the **HCV replicon** without suppressing host cellular gene expression. Synthetic siRNA against NS3-1 also inhibited replication of the **HCV replicon** in a dose-dependent manner. Taken together, the present results imply the possibility that the recombinant lentivirus expressing shRNA against NS3-1 would be a useful tool to inhibit HCV-1b infection.
REFERENCE COUNT: 53 THERE ARE 53 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 4 OF 10 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2004:596261 CAPLUS
DOCUMENT NUMBER: 141:326588
TITLE: Dominant negative effect of wild-type NS5A on NS5A-adapted subgenomic hepatitis C virus RNA replicon
AUTHOR(S): Graziani, Rita; Paonessa, Giacomo
CORPORATE SOURCE: Istituto di Ricerche di Biologia Molecolare P. Angeletti (IRBM), Pomezia, I-00040, Italy
SOURCE: Journal of General Virology (2004), 85(7), 1867-1875
CODEN: JGVIAY; ISSN: 0022-1317
PUBLISHER: Society for General Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English
AB An efficient model is currently used to study hepatitis C virus (HCV) replication in cell culture. It involves transfection in Huh7, a hepatoma-derived cell line, of an antibiotic (neomycin) selectable HCV subgenomic replicon encoding the non-structural (NS) proteins from NS3 to NS5B. However, strong and sustained replication is achieved only on the appearance of adaptive mutations in viral proteins. The most effective of these adaptive mutations are concentrated mainly in NS5A, not only into the original Con1 but also in the recently established HCV-BK and HCV-H77 isolate-derived replicons. This suggests that the expression of wild-type (wt) NS5A may not allow efficient HCV RNA replication in cell culture. With the use of a β-lactamase reporter gene as a marker for HCV replication and TaqMan RNA anal., the replication of different **HCV replicons** in cotransfection expts. was investigated. Comparing wt with NS5A-adapted replicons, the strong evidence accumulated showed that the expression of wt NS5A was actually able to inhibit the replication of NS5A-adapted replicons. This feature was characterized as a dominant neg. effect. Interestingly, an NS5B (R2884G)-adapted replicon, containing a wt NS5A, was dominant neg. on an NS5A-adapted replicon but was not inhibited by the original Con1 replicon. In conclusion, these studies revealed that the original wt Con1 replicon is not only incompetent for replication in cell culture, but is also able to interfere with NS5A-adapted replicons.
REFERENCE COUNT: 33 THERE ARE 33 CITED REFERENCES AVAILABLE FOR THIS

RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 5 OF 10 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2004:169616 CAPLUS
DOCUMENT NUMBER: 140:369763
TITLE: Sequence requirements for the development of a chimeric **HCV replicon** system
AUTHOR(S): Gates, Adam T.; Sarisky, Robert T.; Gu, Baohua
CORPORATE SOURCE: The Metabolic and Viral Diseases Center of Excellence in Drug Discovery, Department of Virology, GlaxoSmithKline Pharmaceuticals, Collegeville, PA, 19426-0989, USA
SOURCE: Virus Research (2004), 100(2), 213-222
CODEN: VIREDF; ISSN: 0168-1702
PUBLISHER: Elsevier Science B.V.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The hepatitis C virus (HCV) 3' nontranslated region (3'NTR) is important for virus infection and replicon replication. Here, the authors constructed a panel of chimera replicons containing non-structural (NS) and 3'NTR sequences from different HCV strains or types, and examined the requirements for stable replication. A subgenomic replicon chimera comprising the polymerase and 3'NTR from HCV strain Con1, and other non-structural genes from type 1a strain H77, supported stable colony formation and replication in Huh7 cells. However, extending the type 1a sequence to include 132 amino acids of NS5B resulted in a defective **HCV replicon**. In contrast, a similar chimera containing HCV strain J4 sequences linked in cis to Con1 NS5B and 3'NTR supported stable replication suggesting that the interaction between the NS proteins and the 3'NTR may represent a critical determinant. Lastly, the type 1a 3'NTR from pCV-J4L6S was unable to confer replication when paired with non-structural coding sequences from BB7 or J4 and the 3'NTR from Con1 was unable to confer replication when paired with J4 or H77 sequences. These results highlighted the importance of sequence specific interaction among 3'NTR and two distinct subdomains of the NS coding region as a determinant in supporting stable replication of subgenomic replicons. The results underscore the importance of directly cloning 3'NTR sequences from relevant clin. samples.
REFERENCE COUNT: 24 THERE ARE 24 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 6 OF 10 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2003:935820 CAPLUS
DOCUMENT NUMBER: 140:156738
TITLE: Characterization of Resistance to Non-obligate Chain-terminating Ribonucleoside Analogs That Inhibit Hepatitis C Virus Replication in Vitro
AUTHOR(S): Migliaccio, Giovanni; Tomassini, Joanne E.; Carroll, Steven S.; Tomei, Licia; Altamura, Sergio; Bhat, Balkrishen; Bartholomew, Linda; Bosserman, Michele R.; Ceccacci, Alessandra; Colwell, Lawrence F.; Cortese, Riccardo; De Francesco, Raffaele; Eldrup, Anne B.; Getty, Krista L.; Hou, Xiaoli S.; LaFemina, Robert L.; Ludmerer, Steven W.; MacCoss, Malcolm; McMasters, Daniel R.; Stahlhut, Mark W.; Olsen, David B.; Hazuda, Daria J.; Flores, Osvaldo A.
CORPORATE SOURCE: Department of Biochemistry, Istituto di Ricerche di Biologia Molecolare P. Angeletti, Pomezia, 00040, Italy
SOURCE: Journal of Biological Chemistry (2003), 278(49), 49164-49170
CODEN: JBCHA3; ISSN: 0021-9258
PUBLISHER: American Society for Biochemistry and Molecular Biology
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The urgent need for efficacious drugs to treat chronic hepatitis C virus (HCV) infection requires a concerted effort to develop inhibitors specific

for virally encoded enzymes. We demonstrate that 2'-C-Me ribonucleosides are efficient chain-terminating inhibitors of HCV genome replication. Characterization of drug-resistant **HCV replicons** defined a single S282T mutation within the active site of the viral polymerase that conferred loss of sensitivity to structurally related compds. in both replicon and isolated polymerase assays. Biochem. analyses demonstrated that resistance at the level of the enzyme results from a combination of reduced affinity of the mutant polymerase for the drug and an increased ability to extend the incorporated nucleoside analog. Importantly, the combination of these agents with interferon-α results in synergistic inhibition of HCV genome replication in cell culture. Furthermore, 2'-C-methyl-substituted ribonucleosides also inhibited replication of genetically related viruses such as bovine diarrhea virus, yellow fever, and West African Nile viruses. These observations, together with the finding that 2'-C-methyl-guanosine in particular has a favorable pharmacol. profile, suggest that this class of compds. may have broad utility in the treatment of HCV and other flavivirus infections.

REFERENCE COUNT: 27 THERE ARE 27 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 7 OF 10 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2003:818518 CAPLUS
DOCUMENT NUMBER: 139:318419
TITLE: Construction of hepatitis C virus sub-genomic replicons and uses for drug screening
INVENTOR(S): Gates, Adam; Gu, Baohua; Sarisky, Robert T.
PATENT ASSIGNEE(S): Smithkline Beecham Corporation, USA
SOURCE: PCT Int. Appl., 159 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003085084	A2	20031016	WO 2003-US10177	20030403
WO 2003085084	A3	20040722		
W: AE, AG, AL, AU, BA, BB, BR, BZ, CA, CN, CO, CR, CU, DM, DZ, EC, GD, GE, HR, HU, ID, IL, IN, IS, JP, KP, KR, LC, LK, LR, LT, LV, MA, MG, MK, MN, MX, NO, NZ, OM, PH, PL, RO, SC, SG, TN, TT, UA, US, UZ, VC, VN, YU, ZA				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
EP 1490389	A2	20041229	EP 2003-723885	20030403
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, SK				

PRIORITY APPLN. INFO.: US 2002-369685P P 20020403
WO 2003-US10177 W 20030403

AB The present invention relates generally to the construction of sub-genomic **HCV replicon** systems that may provide the foundation for generating **HCV replicons** of all six major genotypes and subtypes to facilitate screening, testing, and evaluating anti-infective agents for HCV disease(s). Specifically, the invention relate to nucleotide sequences derived from various functional chimeric **HCV replicons**. The invention describes the successful generation of stable cell lines expressing and replicating functional replicons, containing sequences from HCV genotype 1a (strain H77) or genotype 1b (strain J4) within the prototype 1b replicon backbone from HCV strain BB7.

L4 ANSWER 8 OF 10 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2003:592087 CAPLUS
DOCUMENT NUMBER: 139:360281
TITLE: Alcohol potentiates hepatitis C virus replicon

expression

AUTHOR(S): Zhang, Ting; Li, Yuan; Lai, Jian-Ping; Douglas, Steven D.; Metzger, David S.; O'Brien, Charles P.; Ho, Wen-Zhe

CORPORATE SOURCE: Division of Allergy and Immunology, Joseph Stokes Jr. Research Institute at The Children's Hospital of Philadelphia, Department of Pediatrics, University of Pennsylvania School of Medicine, Philadelphia, PA, USA

SOURCE: Hepatology (Philadelphia, PA, United States) (2003), 38(1), 57-65
CODEN: HPTLD9; ISSN: 0270-9139

PUBLISHER: W. B. Saunders Co.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Alc. consumption accelerates liver damage and diminishes the anti-hepatitis C virus (HCV) effect of interferon alfa (IFN-α) in patients with HCV infection. It is unknown, however, whether alc.. enhances HCV replication and promotes HCV disease progression. The availability of the **HCV replicon** containing hepatic cells has provided a unique opportunity to investigate the interaction between alc. and **HCV replicon** expression. The authors determined whether alc. enhances HCV RNA expression in the replicon containing hepatic cells. Alc., in a concentration-dependent fashion, significantly increased **HCV replicon** expression. Alc. also compromised the anti-HCV effect of IFN-α. Investigation of the mechanism(s) responsible for the alc. action on **HCV replicon** indicated that alc. activated nuclear factor .vkappa.B (NF-.vkappa.B) promoter. Caffeic acid phenethyl ester (CAPE), a specific inhibitor of the activation of NF-.vkappa.B, abolished alc.-induced HCV RNA expression. In addition, naltrexone, an opiate receptor antagonist, abrogated the enhancing effect of alc. on **HCV replicon** expression. In conclusion, alc., probably through the activation of NF-.vkappa.B and the endogenous opioid system, enhances **HCV replicon** expression and compromises the anti-HCV effect of IFN-α. Thus, alc. may play an important role in vivo as a cofactor in HCV disease progression and compromise IFN-α-based therapy against HCV infection.

REFERENCE COUNT: 50 THERE ARE 50 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 9 OF 10 CAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2003:571014 CAPLUS

DOCUMENT NUMBER: 139:129100

TITLE: Construction of GB virus B based replicons and replicon enhanced cells

INVENTOR(S): De Tomassi, Amedeo; Graziani, Rita; Paonessa, Giacomo; Traboni, Cinzia

PATENT ASSIGNEE(S): Istituto Di Ricerche Di Biologia Molecolare P. Angeletti Spa, Italy

SOURCE: PCT Int. Appl., 81 pp.
CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003059944	A2	20030724	WO 2003-EP281	20030113
WO 2003059944	A3	20040205		
W: CA, JP, US				
RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SK, TR				
EP 1468100	A2	20041020	EP 2003-702433	20030113
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, FI, CY, TR, BG, CZ, EE, SK				

PRIORITY APPLN. INFO.: US 2002-348573P P 20020115
US 2002-386655P P 20020606

WO 2003-EP281 W 20030113

AB The present invention features methods for producing GBV-B replicons and replicon enhanced cells. A GBV-B replicon is an RNA mol. able to autonomously replicate in a cultured cell and produce detectable levels of one or more GBV-B proteins. GBV-B replicon enhanced cells are cells having an increased ability to maintain a GBV-B replicon.

L4 ANSWER 10 OF 10 CAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2003:526765 CAPLUS
DOCUMENT NUMBER: 139:377781
TITLE: Nonstructural protein precursor NS4A/B from hepatitis C virus alters function and ultrastructure of host secretory apparatus
AUTHOR(S): Konan, Kouacou V.; Giddings, Thomas H.; Ikeda, Masanori; Li, Kui; Lemon, Stanley M.; Kirkegaard, Karla
CORPORATE SOURCE: Department of Microbiology and Immunology, Stanford University School of Medicine, Stanford, CA, USA
SOURCE: Journal of Virology (2003), 77(14), 7843-7855
CODEN: JOVIAM; ISSN: 0022-538X
PUBLISHER: American Society for Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The nonstructural proteins of hepatitis C virus (HCV) have been shown previously to localize to the endoplasmic reticulum (ER) when expressed singly or in the context of other HCV proteins. To determine whether the expression of HCV nonstructural proteins alters ER function, we tested the effect of expression of NS2/3/4A, NS4A, NS4B, NS4A/B, NS4B/5A, NS5A, and NS5B from genotype 1b HCV on anterograde traffic from the ER to the Golgi apparatus Only the nominal precursor protein NS4A/B affected the rate of ER-to-Golgi traffic, slowing the rate of Golgi-specific modification of the vesicular stomatitis virus G protein expressed by transfection by approx. threefold. This inhibition of ER-to-Golgi traffic was not observed upon expression of the processed proteins NS4A and NS4B, singly or in combination. To determine whether secretion of other cargo proteins was inhibited by NS4A/B expression, we monitored the appearance of newly synthesized proteins on the cell surface in the presence and absence of NS4A/B expression; levels of all were reduced in the presence of NS4A/B. This reduction is also seen in cells that contain genome length **HCV replicons**: the rate of appearance of major histocompatibility complex class I (MHC-I) on the cell surface was reduced by three- to fivefold compared to that for a cured cell line. The inhibition of protein secretion caused by NS4A/B does not correlate with the ultrastructural changes leading to the formation a "membranous web" (D. Egger et al., J. Virol. 76:5974-5984, 2002), which can be caused by expression of NS4B alone. Inhibition of global ER-to-Golgi traffic could, by reducing cytokine secretion, MHC-I presentation, and transport of labile membrane proteins to the cell surface, have significant effects on the host immune response to HCV infection.

REFERENCE COUNT: 81 THERE ARE 81 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT